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ON THE NORMAL AGGLUTININS FOR DIFFERENT KINDS OF PATHOGENIC BACTERIA IN THE SERUM OF COLD-BLOODED ANIMALS

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Normal agglutinins in animal serum for different kinds of bacteria have been studied extensively. Those in the serum of cold-blooded animals, however, seem to have been little investigated. In the literature, I could not find any particular article concerning this problem.

Following Professor Jordan's suggestion, I made a study of the serum of different species of turtle in their content of agglutinins against different kinds of bacteria, especially those which do not grow well at such low temperatures as those of cold-blooded animals.

The species of turtle tested are as follows: *Chrysemys belli*; *Chrysemys elegans*; *Melacoclemmys lesueurii*; *Melacoclemmys (geographica?)*, and *Chelydra serpentina* (snapping turtle).

Two or three of each species were tested carefully. These were supplied by a firm in Chicago, between April 15 and 24, 1918. During the examination they were put in a box containing some tap water in the animal room of this laboratory.

The serum was taken from the arteria carotis without narcosis. Clear serum was separated and used for agglutination with different kinds of bacteria.

The following kinds of bacteria, stock cultures in this laboratory, were tested with the serum of normal turtles:

B. typhosus (Hopkins), *B. paratyphosus* A., No. 4; *B. paratyphosus* B., No. 12; *B. coli communis*; *B. dysenteriae*-Shiga; *B. dysenteriae*-Flexner; *B. fecalis alcaligenes*; *B. pyocyaneus*, and *Staph. aureus* I, II, III, and IV; also *Staph. albus*.

The usual technic of agglutination with small tubes was employed. The titer limit of agglutinins will be indicated in Table 1 by the dilution of serum in the tube in which slightly but decidedly positive agglutination was to be seen macroscopically. Many readings were made at different intervals, but only the one at the 6th hour incubation is noted in the table.

B. typhosus can be agglutinated by the normal serum of different species of turtle at a dilution of 20-60.

B. paratyphosus A can be agglutinated by the serum of *Chrysemys belli* at a dilution of 80.

B. paratyphosus B can be agglutinated by the normal serum of different species of turtle at a dilution of 20-40.

B. coli communis can be agglutinated by 40-80 times dilution of normal serum of *Chrysemys elegans* and *Melacoclemmys lesueurii* and (*geographica?*).

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Shiga dysentery bacilli can be agglutinated by the normal serum of different species of turtle at 30-80 dilution, while Flexner dysentery bacilli are agglutinated by 160-240 dilution of those serums.

The normal serum of snapping turtle (*Chelydra serpentina*) shows a very high agglutinating value for Shiga bacilli (320-640), but a very low titer for Flexner bacilli.

B. fecalis alcaligenes can be agglutinated only slightly by the normal serum of different turtles.

B. pyocyaneus can be agglutinated by the normal serums of different species of turtle at dilution of 40-160. The rate of agglutination reaction of this bacillus is very rapid; usually within half an hour at incubation temperature the agglutination reaches the titer limit.

Different strains of *Staph. aureus* and *albus* can be agglutinated moderately by the normal serum of different genus of *Chrysemys* and *Melacoclemmys* at 20-40, or even 600-1200, times dilution. The serum of snapping turtle (*Chelydra serpentina*) shows a pretty high agglutinating value for several strains of *staphylococcus* (160-320 or even 640 times dilution).

TABLE 1

TESTS MADE WITH STOCK CULTURES OF BACTERIA AND SERUM OF DIFFERENT SPECIES OF TURTLES

	<i>Chrysemys bellii</i>	<i>Chrysemys elegans</i>	<i>Melacoclemmys lesueurii</i>	<i>Melacoclemmys (geographica?)</i>	<i>Chelydra serpentina</i>
<i>B. typhosus</i>	20	20-40	60	20-40	20
<i>B. paratyphosus A</i>	80	40	30	10	20
<i>B. paratyphosus B</i>	40	20	30	20-40	40
<i>B. coli communis</i>	10	40-80	60	80	40-80
<i>B. dysenteriae Shiga</i>	40	40-80	30	80	320-640
<i>B. dysenteriae Flexner</i>	Not tested	160	240	160	20
<i>B. fecalis alcaligenes</i>	10	20	Not tested	10	Not tested
<i>B. pyocyaneus</i>	40-80	80-160	40	20	Not tested
<i>Staph. aureus I</i>	320	1,200	960	1,200	320
<i>Staph. aureus II</i>	Not tested	40	40	40	160
<i>Staph. aureus III</i>	Not tested	20-40	40	10	640
<i>Staph. aureus IV</i>	Not tested	20-40	40	20	320
<i>Staph. albus</i>	Not tested	20-40	40	10	80

As far as I am aware, "normal agglutinins" are not present in new-born warm-blooded animals,¹ and infants show lower normal agglutinating values than adults. It has been shown that living bacteria may enter the lymphatic and portal circulation from the intestine while the individual is apparently in perfect health. According to the prevailing opinion it is a possible and not unreasonable supposition that the presence of "normal agglutinins" is due to slight unrecognized infection with some specific bacilli or to the absorption of specific toxic substances from the intestinal canal.

It would be of considerable interest, therefore, to determine the agglutinating values of the serum of young cold-blooded animals (for

¹ Savage, W. G.: Jour. Hyg., 1918, 17, p. 34.

instance, turtles) at different intervals for different kinds of pathogenic bacteria. Unfortunately, material for this purpose has not been available.

Since I have not tested the serum of very young cold-blooded animals, I am unable to give a definite opinion, but it does not seem probable that those agglutinins in cold-blooded animals for different kinds of pathogenic bacteria, which grow usually with great difficulty at such a low temperature as the body temperature of cold-blooded animals, are due to slight unrecognized infection with some specific bacteria or to the absorption of some toxic substance from the intestinal canal.

CONCLUSIONS

1. The normal serums of *Chrysemys belli*, *Chrysemys elegans*, *Melacoclemmys lesueurii* and *Melacoclemmys* (*geographica*?) show fairly high agglutinating values for certain bacteria which are pathogenic for warm-blooded animals and do not grow readily at the body temperature of cold-blooded animals.

2. These facts are theoretically interesting, because, according to the prevalent hypothesis, the occurrence of normal agglutinins is due to a slight unrecognized infection or to the absorption of some specific toxic substance from the intestinal canal during life. This seems improbable, especially in the case of cold-blooded animals.